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RESEARCH ARTICLE

The interactions of Cr (VI) concentrations and amendments (biochar and manure) on growth and metal accumulation of two species of *Salicornia* in contaminated soil

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Abstract
Heavy metals are among the most dangerous contaminants in the environment. Organic components and plant species that can accumulate and stabilize heavy metals in their organs are a good option for soil remediation of these elements. Therefore, this study aimed to investigate the effects of manure and biochar on the accumulation of heavy metals by *Salicornia* species. *Salicornia persica* Akhani and *Salicornia perspolitana* Akhani were cultivated outdoor in experimental pots. The effects of experimental treatments, including Cr (VI) concentrations, manure, and biochar on the two studied species, were investigated. The results indicated a significant effect ($p < 0.05$) of biochar on the accumulation of heavy metals by two species, *S. persica* and *S. perspolitana*, so that Cr concentrations in the roots and shoots were 258 and 5.41 mg/kg, respectively. In addition, Cr accumulations under manure treatments in the roots and shoots were 334.34 and 9.79 mg/kg, respectively. The content of photosynthetic pigments in both *S. persica* and *S. perspolitana* species under biochar treatment was higher than in control and manure treatments. In general, one can conclude that the accumulation of Cr in *S. perspolitana* was higher than in *S. persica*. Applying biochar and manure amendments could stabilize Cr in soil and reduce Cr accumulation in both *S. persica* and *S. perspolitana* species.

Keywords Soil contamination · *Salicornia* · Phytoremediation · Cr

Introduction

Increased agricultural activities, as well as industrialization in recent decades, have significantly resulted in the accumulation of various contaminants in the environment, especially in soil and water. These contaminants enter the environment through mining operations, discharge of industrial effluents, pesticide-based agriculture, fertilizers, etc. and are challenging and problematic due to harmful effects on soil biological systems (Edao 2017). Removal of heavy metals from soil could be a severe issue as heavy metals are particular contaminants, which, in many cases, could remain in the soil for hundreds (even thousands) of years. Several treatment strategies have been reported to stabilize heavy metals from contaminated soils, including solidification/stabilization (Wang et al. 2021b), soil leaching (Huang et al. 2021), electrokinetic modification (Wang et al. 2021a), oxidation, and chemical reduction (Gao et al. 2021). Despite their efficiency, traditional technologies are costly, time-consuming, and harmful to the environment. In other words, the long-term viability of these traditional practices is severely

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affected by their environmental, social, and economic consequences. More efforts have been made to apply natural solutions such as phytoremediation (Zanganeh et al. 2021), phytomanagement (Zine et al. 2021), microbial-based bioremediation (Zanganeh et al. 2021), and engineered wetland restoration (Yu et al. 2021). Phytoremediation has been one of the “greenest” remediation strategies (Wang et al. 2021b).

Phytoremediation is described as the use of diverse plants to decrease heavy metal concentrations in the soil. Generally, bioremediation is considered cost-effective and environmentally friendly (Yao et al. 2012). During the phytoremediation process, metals are effectively removed from the soil. The efficiency of the phytoremediation process depends on the performance of the plant as well as the effective transfer of metals from the plant roots to the shoots. Plants that accumulate considerable amounts of metal usually have small shoots, so the critical point for increasing the efficiency of the phytoremediation process is how to change the plant biomass to increase metal uptake (Evangelou et al. 2007).

Biochar is a porous carbon product obtained from the pyrolysis of plant-derived organic matter (tree bark, rice husk, pinewood, etc.) or non-plant-derived biomass (cattle manure, poultry manure, etc.). Adding biochar to soil boosts soil biological activity and changes physicochemical properties (pH, CEC, mineral, and organic matter content), resulting in higher crop yields (Bashir et al. 2018; Dhir 2021). Al-Wabel et al. (2015) showed that biochar application improved the growth of maize plants (Zea mays L.) and increased the soil water holding capacity of the soil. Mohamed et al. (2017) also found that biochar addition to heavy metal-contaminated soil increased wheat germination percentage and shoot length. Similar results were obtained by Zanganeh et al. (2021) for the increment in Portulaca oleracea growth grown in contaminated soil. Furthermore, utilizing biochar to promote plant development may influence the fraction and absorption of heavy metals in plants. In other words, biochars are considered to be an excellent alternative for polluted soil remediation (Beesley et al. 2011). Arshad et al. (2017) found that biochar amendment reduced Cr (VI) phytotoxicity and phytoavailability. Kumar et al. (2020a) and Nigussie et al. (2012) demonstrated that adding biochar to soil contaminated with Cr effectively reduced Cr phytotoxicity by lowering the plant’s bioavailability. Plants’ uptake of Cr (VI) is influenced by biochar through two mechanisms: (1) direct adsorption by biochar via electrostatic attraction, complexation, ion exchange, precipitation, and physical adsorption; (2) improving the adsorption capacity of soil particles by changing the physicochemical properties (soil pH, EC, mineral, and organic matter content), followed by biochar adsorption; and (3) decreased Cr (VI) mobility by chemically converting soil Cr (VI) to Cr (III) (Kumar et al. 2020b; O'Connor et al. 2018; Wang et al. 2021a). Moreover, the immobilization effect of biochar may be influenced by its physical and chemical properties and variable and complicated environmental conditions. As a result, much more research is needed to develop a more long-term and practical biochar remediation approach for heavy metal-contaminated soil (O'Connor et al. 2018; Wang et al. 2021a).

In addition to biochar, applying some other materials such as animal manure in soils could also increase the phytoremediation efficiency. Organic fertilizers, especially animal manures, contain large amounts of organic matter compared to chemical ones, which can provide nutrient sources such as nitrogen, phosphorus, and potassium overtime for the plant and improve soil’s physical, chemical, and biological properties. Chen et al. (2021) found that chicken manure promoted the root and shoot lengths and weights of Pharbitis purpurea. Rotkittikhun et al. (2007) described the enhanced growth of Vetiveria zizanioides cultivated in lead-contaminated soil with pig manure. Adding manure to the soil was proposed to reduce the deleterious effects of heavy metals such as Cr (VI) to the soil. Antoniadis et al. (2017), Chen et al. (2021), Liu et al. (2020), Molla et al. (2012), and Singh et al. (2007)’s findings showed that manure appears to have had a favorable effect on Cr (VI). According to the above findings, manure affects Cr (VI) uptake by plants via two mechanisms: (a) lower Cr (VI) mobility due to conversion of Cr (VI) to Cr (III) and the formation of organometallic complexes and (b) better plant absorption ability.

Few plant species, including Sutera fodina (Baker and Brooks 1989), Dicoma niccolifera (Wild 1974), and Leersia hexandra Swartz (Zhang et al. 2007), have been reported to have the ability to accumulate high concentrations of Cr in their tissues. Also, mustard (Brassica juncea (L.)) and sunflower (Helianthus annuus L.) have been reported to accumulate high concentrations of Cr in their tissues (Shahandeh 2000). Coupe and Sallami (2013) compared the ability of three plant species, namely Eucalyptus camaldulensis, Brassica juncea (L.), and Medicago sativum, to uptake copper, zinc, and Cr from the soil. They stated that Eucalyptus camaldulensis had the highest capability to uptake these contaminants from the soil. The above-mentioned plants accumulated a significant quantity of Cr and were categorized as Cr phytoremediators. Furthermore, indigenous plants that are better adapted to grow in a particular place and survive in metal-challenged habitats should be screened to improve the application of phytoremediation in real-world settings (Ali et al. 2013; Mahar et al. 2016). Salicornia, a widespread halophyte and salt-tolerant plant, serves as a suitable plant for phytoremediation of heavy metals due to its characteristic roots and the ability to stabilize metals (Van Oosten et al. 2015).

Halophytes are salt-tolerant plants that naturally evolved in saline soils and have amplified growth at low salt concentrations while decreasing growth at much higher salt concentrations (Barrett-Lennard 2002, Orcutt and Nilsen 2000; Tester and Davenport 2003). They make up roughly 1% of the world's flora and 2% of terrestrial plant species, and they can tolerate salt levels that would harm 90% of other plants (Anjum et al. 2014). Halophytes have a pan-tropical or global distribution and can be found in various habitats, such as pure alkaline semi-deserts, steppes, and mangrove forests (Kapler 2019). They are employed as forage and animal feed, a potential source of edible oil, and a phytodesalination and phytoremediation candidate (Liang et al. 2017; Ventura et al. 2015). Halophytes have a higher potential for phytoremediation of heavy metals (Manousaki and Kalogerakis 2011). Salt tolerance is a trait that has a significant impact on halophytes' high resistance to heavy metals (Wang et al. 2014). There are two approaches to considering halophytes for phytoremediation; (1) for phytostabilization, halophytes can be utilized. Some halophytes have an extensive root system and accumulate heavy metals in their roots, but not in their aerial parts, (2) for phytoextraction, halophytes can be used. Halophytes, which primarily accumulate metals in their aerial parts, are ideal for this application (Wang et al. 2014).

Salicornia L. is a member of the *Amaranthaceae* family of annual hygrohalophytes. Data on 28 different Salicornia species can be found in the database eHALOPH-Halophytes (Santos et al. 2016). It is found throughout the Northern Hemisphere's boreal, temperate, and subtropical regions, including North America, Europe, North and West Africa, the Middle East, Central Asia, and the Far East (Slenzka et al. 2013). As a sustainable crop for the food, medicine, bioenergy, and ecology sectors, this plant offers industrial and environmental applications. Salicornia can grow in a saline environment. It is a halophyte that can tolerate up to 1000 mM of NaCl or more, making it one of the world's saltiest plant species (Volkov 2015). In various studies, it has been described as a leafless plant that grows up to 35 cm tall and is densely branched. Furthermore, the plants have articulated succulent green stems (Cárdenas-Pérez et al. 2021). As a sustainable crop for the food, medicine, bioenergy, and ecology sectors, this plant offers both industrial and environmental applications (Cárdenas-Pérez et al. 2021; Patel 2016). S. persica and S. perspolitana are two species of this family, which are native to Iran. These species grow on hypersaline soil and saline lakes in central Iran. S. persica has glabrous leaves, a dark green flower that turns purplish in fruit, a height of up to 60 (–100) cm, and a canopy diameter of up to 80 cm (Akhani 2008). S. perspolitana is glabrous, utterly prostrate on the ground, intricately and robustly branched from the base, forming a circular habit on the ground, with a canopy of 50–60 cm diameter (Akhani 2008). These species demonstrated high tolerance to soil salinity and petroleum contamination (Abdollahzadeh et al. 2019; Babazadeh et al. 2013; Kaviani et al. 2017, 2019).

Several studies have been carried out on the phytoremediation ability of Salicornia plant species to remove heavy metals such as As (Mesa-Marín et al. 2020; Sharma et al. 2010), Cd (Mesa-Marín et al. 2020; Sharma et al. 2010), Cu (Khalilzadeh et al. 2021; Mesa-Marín et al. 2020), Ni (Kaviani et al. 2019; Khalilzadeh et al. 2021; Mesa-Marín et al. 2020; Sharma et al. 2010), and Pb (Khalilzadeh et al. 2021; Mesa-Marín et al. 2020) other than Cr. However, for the first time, we aimed to investigate the ability of S. persica and S. perspolitana in phytoremediation of Cr-contaminated soils. To do this, the effect of the type of amendments (manure and biochar) and Cr (VI) concentration on the capacity of the two mentioned species to accumulate Cr in their tissues from contaminated soil was studied. In this regard, experimental treatments include Cr (VI) concentrations, manure, and biochar.

Materials and methods

Chemicals

Potassium dichromate, hydrochloric acid, nitric acid, sulfuric acid, gallic acid, Folin–Ciocalteu reagent, methanol, diphenylcarbazide, sodium carbonate, and sodium chloride were purchased from Merck Company.

Physical and chemical properties of soil

The soil samples were taken from a depth of 0 to 40 cm. Because the plowing surface is 0–40 cm and the soil plume will be fully blended across this level, this depth was chosen. Soil samples were collected in the northeastern area of Iran (Mashhad), located between 36° 29′ N and 59° 31′ E. After air drying, the samples were passed through a 2-mm sieve. The soil used in this study was characterized by loamy texture, low salinity (EC of 121.8 μS/cm), alkaline pH (8.14), and organic carbon content of 2.56%. The heavy metal analysis showed that the concentration of Cr (VI) in the soil was 2.91 mg/kg.

Physical and chemical properties of manure and biochar

The completely rotten manure used in this study was prepared by Ferdowsi University of Mashhad, Iran. This study's biochar was produced by pyrolyzing three widely accessible and abundant wood wastes found in northern Iran (*Eucalyptus*, *Populus*, and *Parrotia*). The pyrolysis was carried out at 600 °C at 10 °C/min for 1 h in an argon environment. Before performing the experimental treatments, manure and biochar samples were analyzed using conventional laboratory methods previously applied to measure the soil samples. The percentages of organic carbon, total nitrogen, phosphorus, and potassium in the manure were 56%, 2.95%, 0.62%, and 0.94%, respectively. The values for carbon, hydrogen, nitrogen, oxygen, and sulfur in biochar were 73.41%, 2.71%, 0.37%, 23.51% and 0%, respectively. Biochar had a water holding capacity (WHC) of 92% and particle size of 0.5–1 mm. There was no Cr (VI) in manure and biochar. Biochar and fertilizer were all put through a 2-mm sieve before phytoremediation studies. The pH, EC, and ash content of biochar were 7.7, 0.935 dS/m, and 19.5%, respectively.

Preparation of Cr (VI) contaminated soil

Salicornia habitat's native soil was clay-sandy loam with an EC of 1800 µS/cm. Eight hundred grams of loamy soil, 200 g of sand, and 80 mL of 300 mmol NaCl solution were mixed to create this environment.

The Cr (VI) loads were between 0 and 150 mg/kg of soil. Spiking was done using a diluted solution (2.83 g $K_2Cr_2O_7$ in 1L of deionized water, corresponding to 1000 mg/L Cr (VI) that was added as follows: The unspiked control received no solution; the Cr (VI)-5 treatment received 5 mL of solution per kg of soil; the Cr (VI)-10 treatment received 10 mL per kg of soil; the Cr (VI)-15 treatment received 15 mL per kg of soil; and the Cr (VI)-50, Cr (VI)-100, and Cr (VI)-150 treatments received 50, 100, and 150 mL per kg of soil, respectively. The soil samples were homogenized after treatment and placed into nylons plastic bags. The treated sample was moistened to field capacity and maintained at this moisture level for 15 days. Then, biochar and manure were sieved through a 2-mm sieve and added to the soil samples at 1% w/w (10 g of amendment per 1 kg of soil). The typical percentage assessed in the studies was 1% w/w (Antoniadis et al. 2017). Furthermore, it will not be costly to use in a practical application in terms of cost. The samples, including amendments, were thoroughly blended to ensure that soil conditions were uniform throughout.

Experimental design

The scheme of experimental design was shown in Figs. 1 and 2. The following variables were investigated:

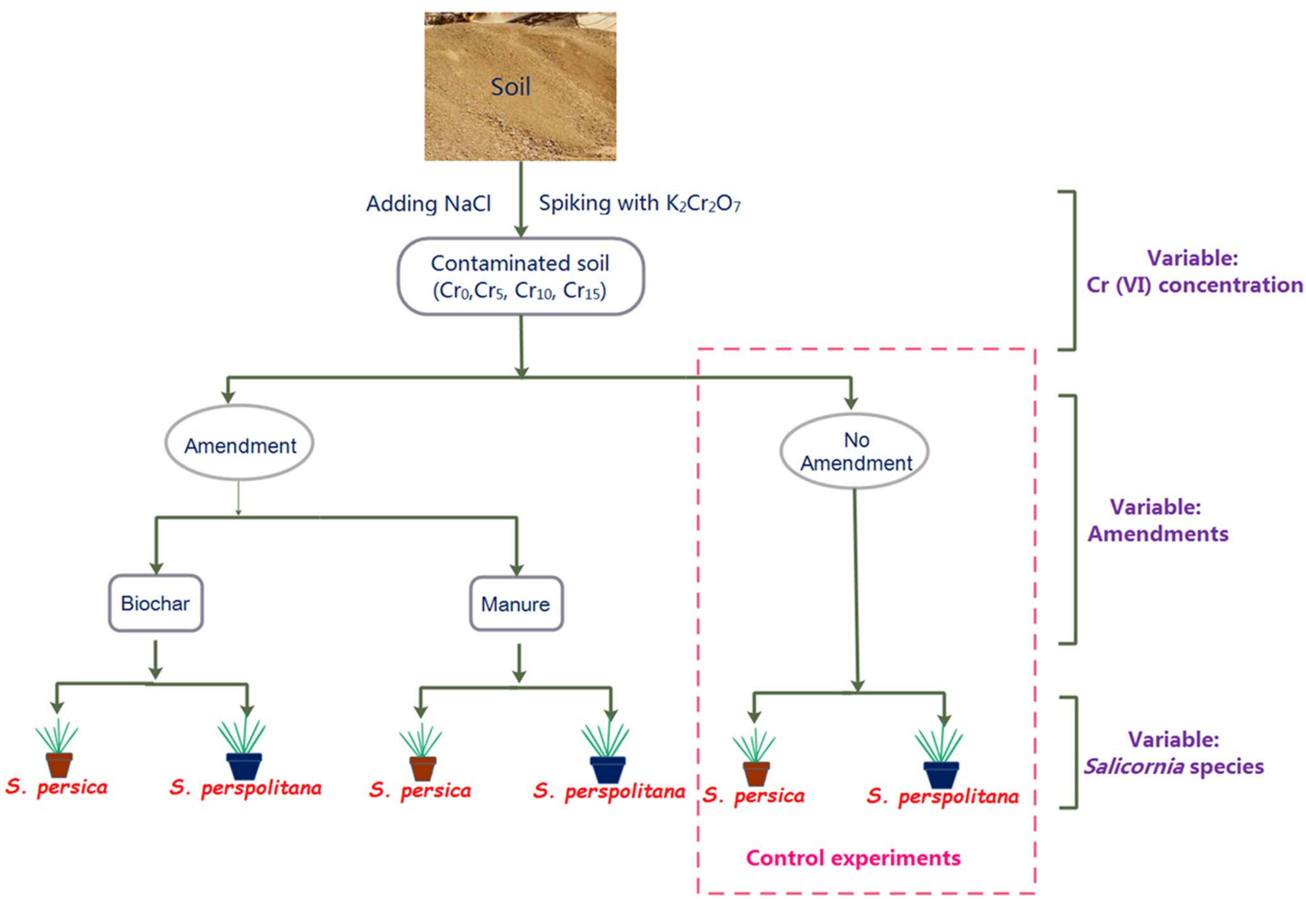

Fig. 1 Experimental set up in this study

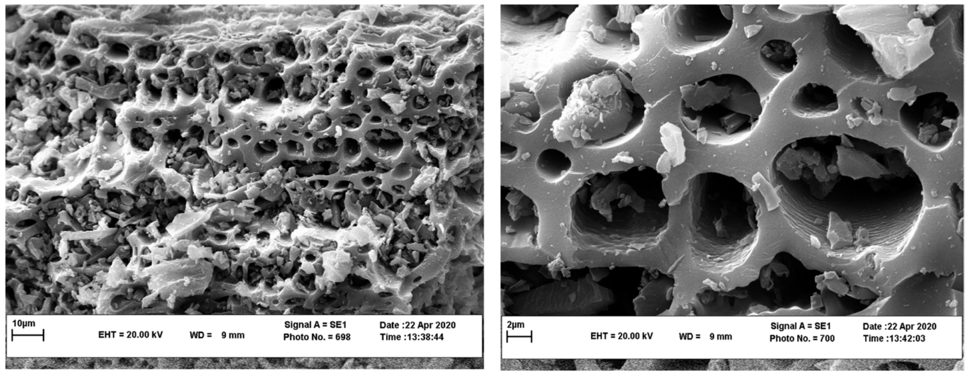

Fig. 2 Scanning electron microscopy (SEM) of biochar surface

i) Four concentrations of Cr (VI) in soil: 0, 5, 10, and 15 mg/kg
ii) Two organic amendments: manure and biochar
iii) Two plant species: *S. persica* and *S. perspolitana*

Controls were soils free of amendments (biochar and manure) and Cr (VI). Two experiment series were established in general: (1) *Salicornia* germination and (2) *Salicornia* phytoremediation. In the first series, the effects of parameters (i) (Cr (VI) concentrations) were researched, whereas, in the second series, the effects of all parameters were investigated.

Germination test

Plant seeds were obtained from the Ala Ava Gene Biotechnology Company in Iran. The Cr was added to the seeds of *S. persica* and *S. perspolitana* species as a factorial design in 3 replicates and seven concentration levels (0, 5, 10, 15, 50, 100, and 150 mg/L). Inside each Petri dish, there were 10 mL of Cr (VI) solution at different concentrations as well as 25 seeds. Then, all the Petri dishes were incubated in a growth chamber at a temperature of 25–30 °C with a photoperiod of 16 h of light and 8 h of darkness. The seeds with a radicle of 2 mm were considered germinated seeds, and all the germinated seeds were counted daily. Because no germination from the 14th to the 16th day, the counting was stopped on the 16th day. Also, the length of root and stem and the length of seedling (the sum of root length and stem length) were measured on the 16th day by an mm-sized ruler. Germination percentage, germination rate, and seedling length were calculated based on the Eqs. 1, 2, 3, 4 and 5 respectively (Saberi et al. 2010).

$$\mathrm{GP} = \frac{\mathrm{G}}{\mathrm{N}} \times 100 \tag{1}$$

$$\mathrm{GS} = \Sigma \frac{\mathrm{ni}}{\mathrm{Di}} \tag{2}$$

$$\mathrm{TL} = \mathrm{SL} + \mathrm{RL} \tag{3}$$

$$\mathrm{SVI} = \frac{\mathrm{GR} \times \mathrm{Mean(SI + RL)}}{100} \tag{4}$$

$$\text{Allometry} = \frac{\text{root length}}{\text{shoot lrngth}} \tag{5}$$

where *GP* is the germination percentage, *G* is the final number of germinated seeds, *N* is the number of seeds sown (25 seeds in this study), *GS* is the germination rate, ni is the number of germinated seeds on counting days, *Di* is the number of days in the experiment, *TL* is the total seedling length, *SVI* is the seed vigor index, *SL* is the stem length, and *RL* is the root length.

Phytoremediation test

A set of plastic pots with a total volume of 2 L (area of 133 cm^2 and depth of 10 cm) was filled with the prepared soil samples. 12 seeds of *Salicornia* were planted at a depth of 0.5 cm for each pot. To ensure the health of the roots, drainage holes were drilled into the bottom of the pots. Seed germination was not observed in the contaminated soil after a month. As a result, seedlings of *S. persica* and *S. perspolitana* were planted in a mixture of cocopeat and perlite (1:1) in a growth chamber at 30 ± 5 °C with photoperiodic lighting (16 h of light: 8 h of dark). In the early stages of the seedlings' growth, Hoagland nutrient solution was used every 2 weeks. After 5 months, seedlings of the same size were selected and transferred to the contaminated soil in the pots. The pots were placed in the outdoor environment. The outdoor experiments lasted 6 months, from May through October 2019. During this time, the average night and day temperatures were 14.14 and 26.75 °C, respectively. The average humidity level was 36.57% (data from the Department of Water Science and Engineering-Ferdowsi University of Mashhad, Iran).

The seedlings died at 50, 100, and 150 mg/kg but survived at 0, 5, 10, and 15 mg/kg concentrations. The seedlings of *S. persica* and *S. perspolitana* were harvested after 6 months of growth in the contaminated soil. After harvesting, the roots and shoots were washed first with tap water and then with distilled water. The fresh and dried weights of the plants' roots and shoots were measured in g/kg/pot.

Measurement of heavy metal concentrations

Soil, fertilizer, and biochar

The concentration of heavy metals in soil, fertilizer, and biochar was determined using the method described by Alghanmi et al. (2015). First, 1 g of sample was mixed with a 3:1 mixture of hydrochloric acid 37% and nitric acid 70% and stirred for 12 h at room temperature. It was then heated for 3 h at 130 °C. The resulting solution was filtered using filter paper. The concentration of hexavalent chromium in the solution was measured using a UV–Visible spectrophotometer (HACH, DR 5000, USA) at 540 nm with diphenylcarbazide as a reagent.

Plant

The method of Antoniadis et al. (2018) was used to determine the amount of chromium in plant tissues. The dried plant parts, including shoots and roots, were crushed and then burned at 500 °C in a furnace for 5 h. After that, 1 g of dried plant sample was mixed with 20 mL of 20% hydrochloric acid and heated to 70 °C for 7 h to complete the digestion of the samples. Finally, the samples were filtered through Whatman filter paper with a pore size of 45 microns. Total Cr contents were determined by inductively coupled plasma spectroscopy (ICP-OES) (model ICP-OES, SPECTRO ARCOS-76004555). The concentration of Cr (VI) in the samples was analyzed using a UV–Visible spectrophotometer (HACH, DR 5000, USA). The difference between Cr (VI) and total Cr was used to calculate the amount of Cr (III).

Measurement of photosynthetic pigments

The content of chlorophyll a and b, as well as carotenoids, was measured using the (Arnon 1967) method. First, 0.5 g of fresh plant material was placed into a porcelain mortar and then crushed and ground thoroughly using liquid nitrogen. Second, the sample was centrifuged at 6000 rpm for 10 min after being soaked in 20 mL of acetone 80%. Third, the sample was then passed through filter paper for further transparency. Finally, the absorbance of the obtained extract was measured UV–Visible spectrophotometer (HACH, DR 5000, USA) at 663 nm for chlorophyll a, 645 nm for chlorophyll b, and 470 nm for carotenoids. The amount of chlorophyll a and b, as well as carotenoids (in mg/g fresh weight), was calculated using the following formulas:

$$\text{Chlorophyll a} = (19.3 \times A663 - 0.86 \times A645)V/100W \tag{6}$$

$$\text{Chlorophyll b} = (19.3 \times A645 - 3.6 \times A663)V/100W \tag{7}$$

$$\text{Carotenoides} = 100(A470) - 3.27(\text{mgChl.a}) - 104(\text{mgChl.b})/227 \tag{8}$$

where V denotes the volume of filtered solution; A denotes light absorption at wavelengths of 663, 645, and 470 nm; and W is the sample's wet weight in grams.

Measuring the total phenolic compounds

The Folin-Ciocalteu method, described by Levizou et al. (2019a), was used to determine the concentration of total phenolic compounds in plant samples. First, the plant's dry above-ground parts were separated and dried at 70 °C for 24 h. Then, a total of 0.2 g of crushed dry plant sample was extracted for 1 h at 40 °C in water and methanol (1:1, v/v). After that, 3.95 mL of deionized water was added to 0.05 mL of extract. Next, 0.25 mL of 10% Folin-Ciocalteu reagent was applied, followed by 0.75 mL of 20% sodium carbonate after 5 min. The adsorption of the mixture was measured at 760 nm after 2 h using a UV–Visible spectrophotometer. The calibration curve was created using gallic acid as the standard. The total phenol content was expressed as gallic acid equivalents per g of dry matter.

Sample analysis

Soil texture was characterized by the hydrometric method using a soil texture triangle. Soil pH and EC (electrical conductivity) were measured using pH-meter (20 +, Crison, Spain) and EC-meter (4510, Jenway, England) devices, respectively. Soil organic carbon was determined by the dry combustion method (Park et al. 2017). The Kjeldahl method with titration was used to determine total nitrogen. Soil available phosphorus was measured according to the Olsen method (1954) using a spectrophotometer (model DR 5000) at the wavelength of 660 nm. Available potassium was determined using the ammonium acetate method and photoelectric flame photometer (Jenway PFP7). Total Cr contents were determined by the inductively coupled plasma spectroscopy (ICP-OES) (model ICP-OES, SPECTRO ARCOS-76004555) and the percentages (%) of carbon, nitrogen, hydrogen, oxygen, and sulfur in the biochar using elemental analysis (FLASH EA 1112 SERIES, Thermo Finnigan). The total phenolic content of the samples was calculated as gallic acid equivalent (mg of gallic acid per g of dry matter). Scanning electronic microscopy (SEM) (Model VP 1450, LEO, Germany) was applied to determine the morphology of the materials used in this study. Experimental treatments included plant species (2 levels), the addition of metal Cr (VI) in 7 levels (zero, 5, 10, 15, 50, 100, and 150 mg/kg), and soil amendments in 3 levels (manure, biochar, and control).

Determination of translocation factor, bioconcentration factor, and biological accumulation coefficient indices

After identifying heavy metal concentrations in plant and soil samples, it should be necessary to calculate indices to evaluate the phytoremediation capability of plants. Translocation factor (TF), bioconcentration factor (BCF), and biological accumulation coefficient (BAC) are potential phytoremediation metrics. The ratio of metal concentrations in shoots to roots is called TF, the ratio of metal concentrations in roots to soil is called BCF, and the ratio of metal concentrations in shoots to soil is called BAC. The TF values greater than 1 indicate that the plant is suitable for extracting the contaminants. Also, plants with TF values and BAC greater than one are suitable for phytoremediation of the contaminants. Plants with a TF value of less than one and a BCF value of more than one are good candidates for phytostabilization (Yoon et al. 2006).

Statistical analysis

This study used a completely randomized design (factorial) to test the effects of different concentrations of Cr (VI) and two amendments (manure and biochar), on the growth of two plant species and heavy metals accumulation in contaminated soil. The total number of experimental units (number of pots) was 126. The data were analyzed using Minitab 16 software, and the means were compared using ANOVA and Turkey's test (*$p < 0.05$*).

Results

Biochar morphology

As shown in scanning electron microscopy (SEM) images (Fig. 1), biochar is characterized by a large surface area and a relatively regular network of honeycomb-shaped pores on its surface, which leads to the absorption of heavy metals. This honeycomb network represents a carbon skeleton in the biochar structure (Ghani et al. 2013). The surface functional groups of biochar were described in our previous study (Zanganeh et al. 2021).

Germination test

The effect of Cr (VI) concentrations on germination traits

Table 1 shows the impact of different Cr (VI) concentrations on germination percentage and rate, root and shoot lengths, and seedling length in the two species studied. For both species, as the concentration of Cr (VI) increased from 0 to 150 mg/L, all germination features decreased significantly, eventually resulting in plant death. *S. persica* has stronger resistance to Cr (VI) stress than *S. perspolitana*, as demonstrated in this table.

Table 1 The effect of different concentrations of Cr on germination traits of *S. persica* and *S. perspolitana* species

Species	Cr (VI) concentration (mg/kg)	Germination percentage	Germination rate	Stem length (cm)	Root length (cm)	Seedling length (cm)
S. persica	0	73.33±2.30^{a}	1.30±0.41^{a}	12.16±0.78^{a}	1.83±0.14^{a}	14.00±0.91^{a}
	5	70.66±2.30^{ab}	1.26±0.41^{ab}	12.15±0.90^{a}	1.51±0.63^{a}	13.73±1.62^{ab}
	25	65.33±2.30^{abc}	1.16±0.41^{abc}	10.80±0.44^{ab}	0.92±0.66^{b}	11.73±0.50^{bcd}
	50	54.66±2.30^{ef}	0.97±0.41^{ef}	10.06±0.11^{bc}	0.86±0.11^{b}	10.93±0.11^{cde}
	100	49.33±6.11^{fg}	0.88±0.10^{fg}	9.15±0.25^{bcd}	0.71±0.28^{b}	9.86±0.23^{def}
	150	40.00±4.00^{hi}	0.71±0.071	6.32±0.11^{fg}	0.67±0.11^{b}	7.00±0.00^{hi}
S. perspolitana	0	64.00±0.0^{bcd}	1.14±0.00^{bcd}	10.60±0.52^{ab}	2.00±0.00^{a}	12.60±0.52^{abc}
	5	58.66±2.30^{cde}	1.04±0.41^{cde}	10.42±1.12^{ab}	1.50±0.11^{a}	11.93±1.13^{abcd}
	25	56.00±0.00^{def}	1.00±0.00^{def}	8.40±0.60^{cde}	0.90±0.00^{b}	9.30±0.60^{efg}
	50	50.66±2.30^{ef}	0.90±0.41^{ef}	7.71±0.51^{def}	0.85±0.00^{b}	8.56±0.51^{fgh}
	100	41.33±2.30^{gh}	0.73±0.41^{gh}	6.91±0.40^{ef}	0.74±0.54^{b}	7.66±0.41^{gh}
	150	32.00±4.00^{i}	0.57±0.71^{i}	4.92±0.71^{g}	0.48±0.34^{b}	5.40±0.69^{i}

Different letters denote statistically significant variations between the means of the control and treated soils at $p > 0.05$

Phytoremediation test

The effect of manure, biochar, and Cr (VI) concentration on the plant growth

As seen in Table 2, the effect of different concentrations of Cr (VI) on the plant height of the two studied species was statistically significant ($p < 0.05$). The highest and lowest heights were related to Cr0 and Cr15, with corresponding heights of 45.33 and 27 cm, respectively. The height reported for *S. persica* was 6.42% higher than *S. perspolitana*, which can be attributed lower accumulation of Cr (VI) in *S. persica* tissues. As shown in the table, the effect of different amendments on the height of the two studied plant species was also significant. Manure and biochar treatments increased height by 22.42% and 12.51%, respectively, compared to the control treatment. Besides, when the Cr (VI) concentration in the soil was increased from zero to 15 mg/kg, the total biomass of both plants decreased considerably. Furthermore, adding biochar and manure to the soil increased the plant's overall biomass. In the absence of amendments, the ratio of shoot to root increased for both species when Cr (VI) concentration rose from 0 to 10 mg/kg and then reduced as the concentration increased to 15 mg/kg. This trend also rose significantly when the content of Cr (VI) was increased and biochar was added to the soil; however, it did not change significantly when manure was applied.

The effect of manure, biochar, and Cr (VI) concentration on metal accumulation

Figure 3 shows the effects of different concentrations of Cr (VI), manure, and biochar on the metal accumulation in the roots of *S. persica* and *S. perspolitana* after 6 months of growing in the contaminated soil. Generally, the accumulation of Cr in the roots of both species was significantly affected by the Cr content in the soil. In other words, the accumulation of Cr in the roots of both species increased by increasing the Cr concentration from 0 to 15 mg/kg in the soil. In the absence of amendment, the highest and lowest accumulations of total Cr, Cr (VI), and Cr (III) was found in the roots of plants grown in soil with zero and 15 mg/kg

Table 2 The effects of manure, biochar, and Cr (VI) concentrations on the height and the weight of *S. persica* and *S. perspolitana*

Species	Amendment	Cr (VI) concentration	Plant height (cm)ns	Dry shoot weight (g per pot)**	Dry root weight (g per pot)	Total biomass(g per pot)**	Shoot/root*
S. persica	-	0	43.6 ± 3.5^{abcde}	1.5 ± 0.2^{c}	0.26 ± 0.01^{b}	1.8 ± 0.2^{b}	5.7 ± 0.6^{c}
		5	32.3 ± 2.5^{fgh}	$0.7\pm0.1d^{e}$	0.08 ± 0.0^{de}	0.8 ± 0.1^{de}	9.1 ± 1.9^{c}
		10	32.6 ± 1.5^{fgh}	0.5 ± 0.1^{def}	0.027 ± 0.0042^{ghi}	0.59 ± 0.09^{def}	21.0 ± 7.5^{ab}
		15	21.33 ± 2.30^{i}	0.18 ± 0.05^{i}	0.014 ± 0.002^{hi}	0.19 ± 0.44^{i}	13.3 ± 4.3^{abc}
	Biochar	0	46.6 ± 2.1^{abc}	1.8 ± 0.09^{ab}	0.27 ± 0.008^{b}	2.09 ± 0.09^{a}	6.6 ± 0.3^{c}
		5	36.5 ± 4.5^{cdefg}	$1.3\pm0.1c$	0.087 ± 0.01^{d}	1.44 ± 0.16^{c}	15.6 ± 1.9^{abc}
		10	33.3 ± 3^{efgh}	0.5 ± 0.05^{defgh}	0.064 ± 0.009^{hi}	0.57 ± 0.037^{defg}	8.1 ± 1.8^{c}
		15	27.6 ± 1.5^{ghi}	0.4 ± 0.07^{fghi}	0.018 ± 0.006^{hi}	0.40 ± 0.07^{fghi}	22.5 ± 7.7^{a}
	Manure	0	48 ± 2^{a}	1.9 ± 0.08^{a}	0.3 ± 0.03^{a}	2.23 ± 0.11^{a}	6.2 ± 0.3^{c}
		5	46 ± 3.1^{abc}	1.5 ± 0.01^{bc}	0.12 ± 0.017^{c}	1.69 ± 0.17^{bc}	12.5 ± 1.3^{abc}
		10	41.3 ± 3.2^{abcdef}	0.7 ± 0.06^{d}	0.08 ± 0.019^{de}	0.84 ± 0.05^{d}	9.7 ± 2.9^{c}
		15	29.6 ± 2.5^{ghi}	0.3 ± 0.02^{fghi}	0.06 ± 0.003^{defg}	0.42 ± 0.02^{fghi}	6.2 ± 0.04^{c}
S. perspolitana	-	0	41.3 ± 2.5^{abcdef}	0.4 ± 0.03^{fghi}	0.050 ± 0.004^{efgh}	0.46 ± 0.04^{fghi}	8.1 ± 0.5^{c}
		5	35 ± 1^{defg}	0.3 ± 0.03^{fghi}	0.035 ± 0.01^{fghi}	0.38 ± 0.04^{fghi}	10.8 ± 3.5^{bc}
		10	23.6 ± 3.5^{hi}	0.26 ± 0.01^{ghi}	0.018 ± 0.005^{hi}	0.28 ± 0.01^{hi}	16 ± 6.7^{abc}
		15	24.3 ± 2.1^{hi}	0.21 ± 0.01^{i}	0.018 ± 0.004^{hi}	0.23 ± 0.01^{i}	12.2 ± 2.7^{abc}
	Biochar	0	45.33 ± 0.57^{abcd}	0.49 ± 0.01^{efgh}	0.06 ± 0.004^{defg}	0.55 ± 0.01^{efgh}	8.3 ± 0.5^{c}
		5	36.6 ± 1.5^{bcdefg}	0.37 ± 0.09^{fghi}	0.038 ± 0.18^{fghi}	0.41 ± 0.11^{fghi}	10.4 ± 2.2^{bc}
		10	32 ± 6.2^{fgh}	0.28 ± 0.02^{ghi}	0.02 ± 0.0009^{hi}	0.30 ± 0.02^{ghi}	12 ± 1.3^{c}
		15	28 ± 3.4^{ghi}	0.25 ± 0.01^{hi}	0.018 ± 0.007	0.27 ± 0.02^{i}	15.3 ± 6.1^{abc}
	Manure	0	47 ± 1^{ab}	0.52 ± 0.03^{defg}	0.06 ± 0.005^{def}	0.58 ± 0.03^{def}	8.1 ± 0.2^{c}
		5	40.3 ± 4.9^{abcdef}	0.51 ± 0.02^{defg}	0.06 ± 0.007^{defg}	0.57 ± 0.02^{defg}	8.3 ± 0.7^{c}
		10	31 ± 7.2^{fghi}	0.33 ± 0.02^{fghi}	0.04 ± 0.006^{efghi}	0.38 ± 0.02^{fghi}	7.1 ± 0.9^{c}
		15	28 ± 3.6^{ghi}	0.37 ± 0.09^{fghi}	0.03 ± 0.002^{fghi}	0.41 ± 0.08^{fghi}	9.7 ± 2.7^{c}

Different letters denote statistically significant variations between the means of the control and treated soils at $p > 0.05$. Asterisk denotes the significance of each factor's interaction effect (species × amendment × concentration); *, **: $p < 0.05$ and 0.01 respectively, *n.s.* not significant

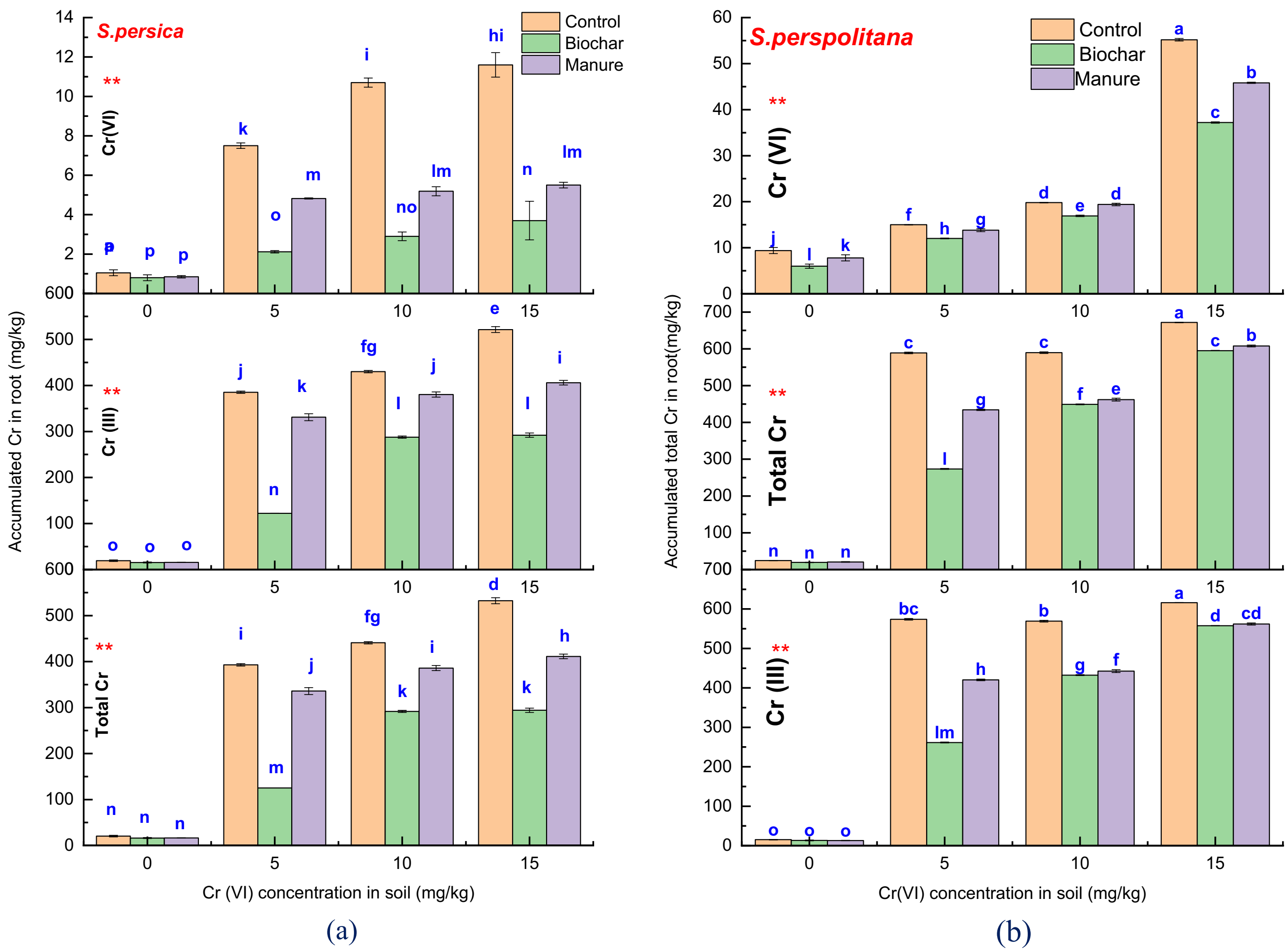

Fig. 3 Effects of manure, biochar, and Cr (VI) concentration on accumulation of various forms of Cr in the root of S. persica and S. perspolitana species. Error bars represent the standard errors of the means. Different letters denote statistically significant variations between the means of the control and treated soils at $p > 0.05$. Asterisk denotes the significance of each factor's interaction effect (species × amendment × concentration); **: $p < 0.01$

metal, respectively. Furthermore, total Cr, Cr (VI), and Cr (III) accumulation in the roots of *S. perspolitana* was higher than in *S. persica*.

The organic compounds used in this study showed a negative effect on the accumulation of total Cr and Cr (III) in the roots of the two studied species. In biochar treatment, the accumulation of total Cr, Cr (VI), and Cr (III) in *S. persica* decreased by 68.12%, 71.9%, and 68.25% at 5 mg/kg; 33.88%, 73.09%, and 93.31% at 10 mg/kg; and 44.65%, 63.10%, and 43.99% at 15 mg/kg. In *S. perspolitana* under biochar treatment, the accumulation of total Cr, Cr (VI), and Cr (III) decreased by 53.54%, 36.10%, and 54.4% at 5 mg/kg; 23.80%, 19.96%, and 24.0% at 10 mg/kg; and 11.39%, 14.54%, and 9.50% at 15 mg/kg. In addition to, in the manure treatment, the accumulation of total Cr, Cr (VI), and Cr (III) in *S. persica* decreased by 14.48%, 36.24%, and 14.07% at 5 mg/kg; 12.49%, 51.85%, and 91.15% at 10 mg/kg; and 22.73%, 52.32%, and 22.13% at 15 mg/kg. In *S. perspolitana* under biochar treatment, the accumulation of total Cr, Cr (VI), and Cr (III) decreased by 26.27%, 16.93%, and 22.96% at 5 mg/kg; 21.63%, 8.11%, and 22.22% at 10 mg/kg; and 9.48%, 2.02%, and 8.81% at 15 mg/kg.

Figure 4 indicates the effects of different concentrations of Cr (VI), manure, and biochar on the accumulation of total Cr, Cr (VI), and Cr (III) in the shoots of *S. persica* and *S. perspolitana* after 6 months of growing in the contaminated soil. Like the results obtained for the roots, the accumulation of Cr in the shoots of both species was significantly affected by the Cr content in the soil. In other words, Cr accumulation in the shoot showed an increasing trend with increasing Cr concentration from 0 to 15 mg/kg in the soil for both species. In *S. persica*, at a Cr concentration of 0 mg/kg, the accumulation of total Cr, Cr (VI), and Cr (III) in the shoots of plants grown in contaminated soil were 2.07, 0.65, and 1.41 mg/kg. At 15 mg/kg, the corresponding values for

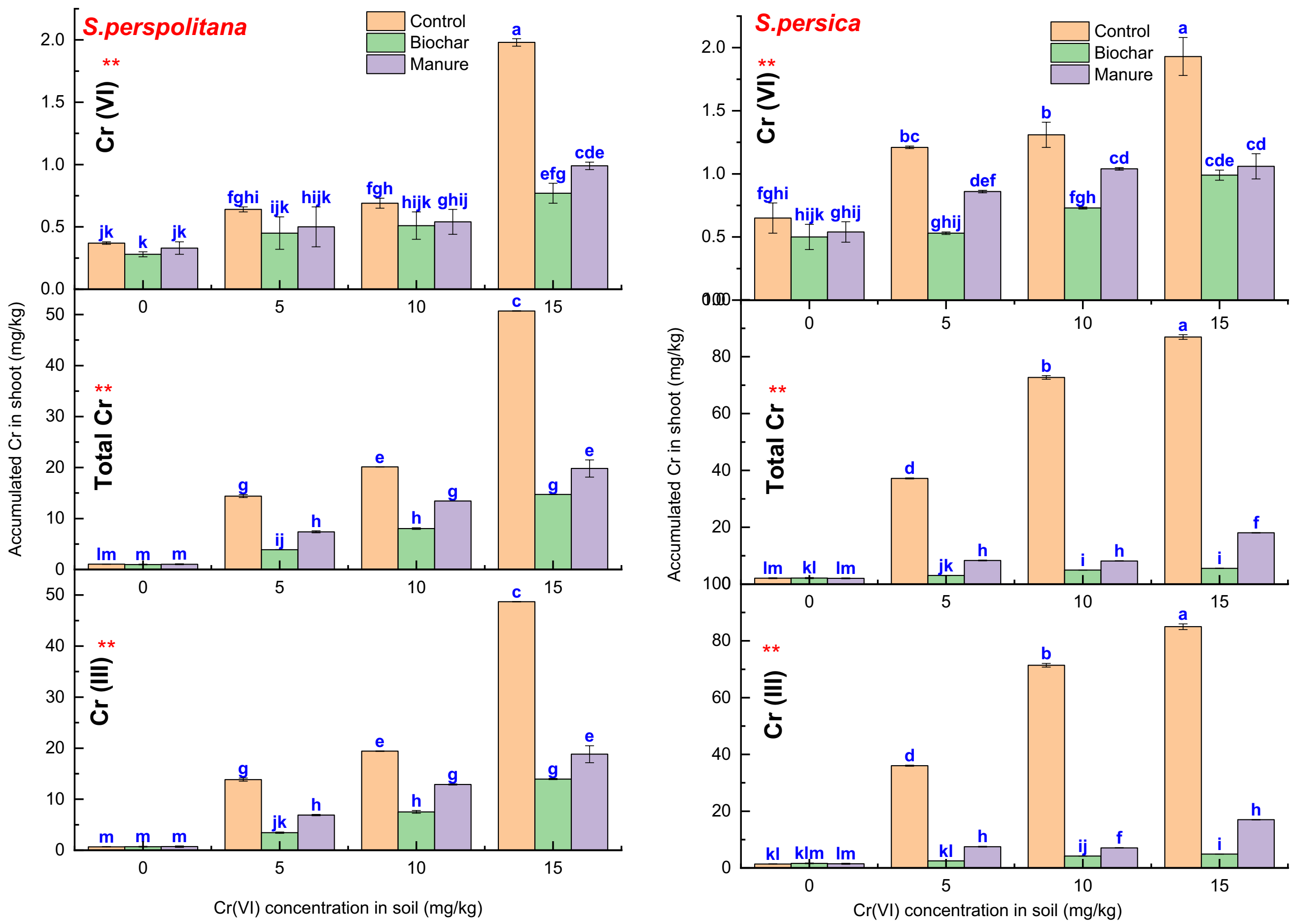

Fig. 4 Effects of different concentrations of Cr (VI), manure, and biochar on accumulation of various froms of Cr in shoots of *S. persica* and *S. perspolitana* species. Error bars represent the standard errors of the means $p > 0.05$. Asterisk denotes the significance of each factor's interaction effect (species × amendment × concentration); $^{**}p < 0.01$

accumulation of total Cr, Cr (VI), and Cr (III) were 86.93, 1.93, and 85.00 mg/kg for this species.

In *S. perspolitana*, at 0 mg/kg, the accumulation of total Cr, Cr (VI), and Cr (III) in the shoots of plants grown in contaminated soil were 1.03, 0.33, and 0.7 mg/kg. At 15 mg/kg, the values for accumulation of total Cr, Cr (VI), and Cr (III) were 50.70, 1.98, and 48.71 mg/kg for this species.

Unlike roots, the accumulation of total Cr, Cr (VI), and Cr (III) in shoots of S. persica were higher than S. perspolitana. As shown in Fig. 4, the organic compounds used in this study hurt the accumulation of total Cr, Cr (VI), and Cr (III) in the shoots of the two species.

In biochar treatment, the accumulation of total Cr, Cr (VI), and Cr (III) in S. persica decreased by 91.83%, 56.19%, and 93.05% at 5 mg/kg; 93.22%, 44.27%, and 94.11% at 10 mg/kg; and 93.60%, 48.70%, and 94.23% at 15 mg/kg. In S. perspolitana under biochar treatment, the accumulation of total Cr, Cr (VI), and Cr (III) decreased by 73.06%, 29.68%, and 75.07% at 5 mg/kg; 60.05%, 26.08%, and 61.29% at 10 mg/kg; and 70.96%, 61.11%, and 71.36% at 15 mg/kg.

In the manure treatment, the accumulation of total Cr, Cr (VI), and Cr (III) in S. persica decreased by 77.71%, 28.92%, and 79.17% at 5 mg/kg; 75.18%, 20.61%, and 76.18% at 10 mg/kg; and 90.62%, 45.07%, and 91.61% at 15 mg/kg. In S. perspolitana under biochar treatment, the accumulation of Cr, Cr (VI), and Cr (III) decreased by 48.89%, 21.87%, and 50.14% at 5 mg/kg; 33.23%, 21.73%, and 33.65% at 10 mg/kg; and 60.90%, 50%, and 61.36% at 15 mg/kg.

Effect of manure, biochar, Cr (VI) concentrations on chlorophyll a and b, carotenoid content, and phenolic content

The effects of different concentrations of Cr (VI), manure, and biochar on the content of photosynthetic pigments in *S. persica* and *S. perspolitana* are shown in Fig. 5. According to these results, different concentrations of Cr (VI) negatively influenced the content of chlorophyll *a* and *b* in the two studied species. Chlorophyll a decreased 27.45%, 28.23%, and 31.37% in *S. persica* at Cr (VI) concentrations of 5, 10, and

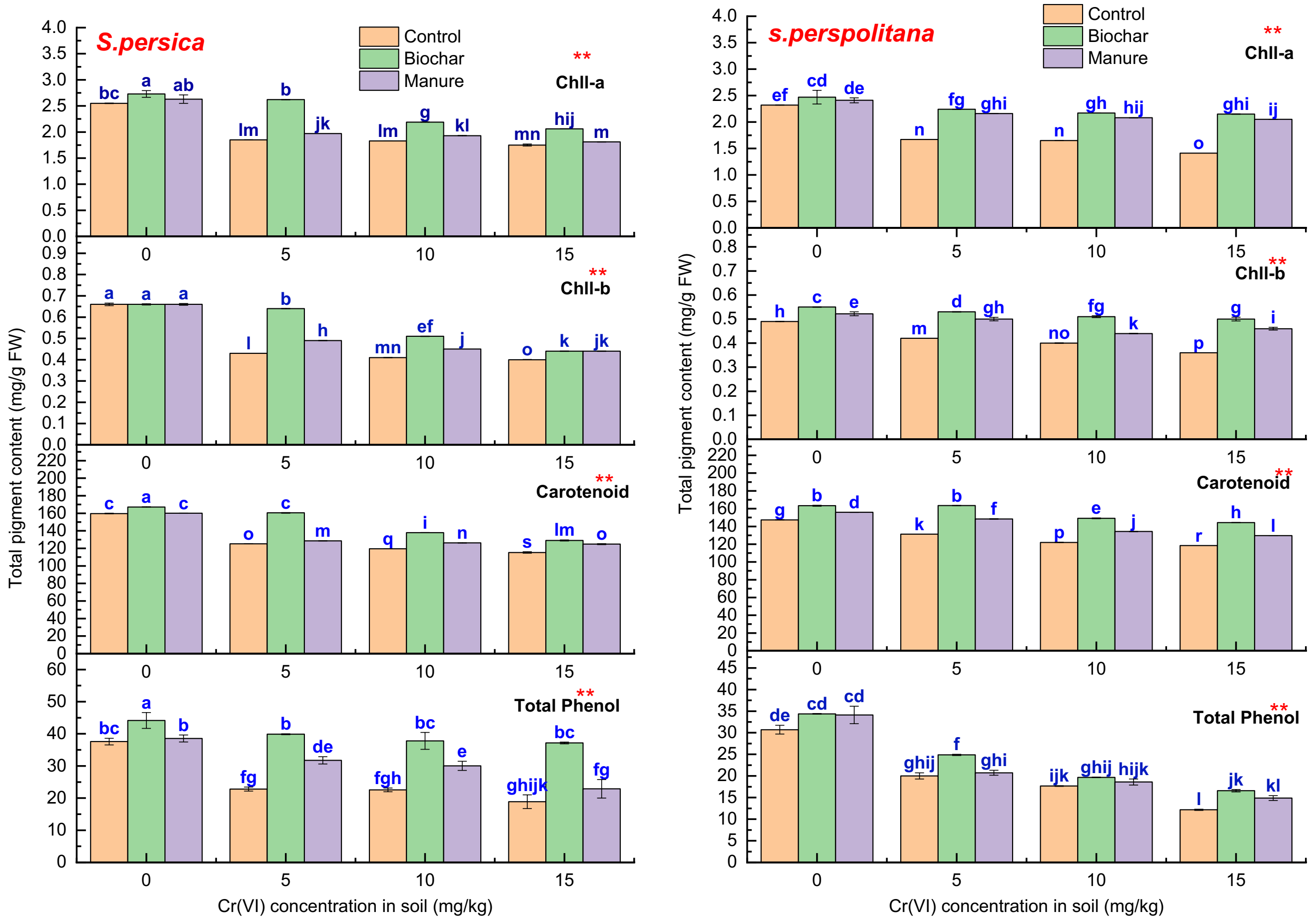

Fig. 5 Effects of manure, biochar, and Cr (VI) concentration on chlorophyll a and b, carotenoid content, and phenolic content. Error bars represent the standard errors of the means $p > 0.05$. Asterisk denotes the significance of each factor's interaction effect (species × amendment × concentration); $^{**}p < 0.01$

15 mg/kg, while chlorophyll b decreased 39.54%, 37.67%, and 39.48%. Chlorophyll content fell by 28.01%, 28.8%, and 39.2% in *S. perspolitana* and chlorophyll b content declined by 28%, 28.8%, and 39.2%, respectively, as compared to controls. The application of Cr (VI) resulted in a considerable decrease in chlorophyll concentration. Regarding declines in chlorophyll a and b, there was a substantial difference between different Cr (VI) concentrations (Fig. 5).

The organic compounds (manure and biochar) used in this study also indicated a positive effect on the content of photosynthetic pigments in the two studied species. Chlorophyll content increased by 41.6%, 19.6%, and 17.7%, while chlorophyll b levels increased by 50.9%, 25.4%, and 11.2%, respectively, when *S. persica* species were treated with biochar at Cr (VI) concentrations of 5, 10, and 15 mg/kg. When biochar was applied to *S. perspolitana*, it increased chlorophyll a by 34.1%, 31.5%, and 52.48% and chlorophyll b by 26.85%, 27.5%, and 40.66%, respectively, compared to the controls (without amendment). In *S. persica* species, the content of chlorophyll a fell by 6.4%, 5.4%, and 3.4%, while the content of chlorophyll b was reduced by 13.9%, 10.19%, and 11.7%, respectively, in the manure treatment at Cr (VI) concentrations of 5, 10, and 15 mg/kg. Compared to the control, declines in chlorophyll a were 29.3%, 26.06%, and 45.3%, and decreases in chlorophyll b were 18.82%, 11%, and 29.9% for *S. perspolitana* species (without amendment).

In general, the highest content of chlorophyll *a* (2.73 mg/g fresh weight) was found in *S. persica* species at a Cr (VI) concentration of 0 mg/kg in biochar treatment, and the lowest content (1.41 mg/g fresh weight) was found for *S. perspolitana* species at Cr (VI) concentration of 15 mg/kg in the control treatment (without amendment). Also, the highest content of chlorophyll *b* (0.669 mg/g fresh weight) was found in *S. persica* species at Cr (VI) concentration of 0 mg/kg in biochar treatment, and the lowest content (0.36 mg/g fresh weight) has belonged to *S. perspolitana* species at Cr (VI) concentration of 15 mg/kg in the control treatment (without amendment).

Figure 5 shows the results of the effects of different concentrations of Cr (VI), manure, and biochar on the carotenoid content of *S.persica* and *S. perspolitana*. According to these results, the different concentrations of Cr (VI) negatively influenced the carotenoid content in the two species. In *S. persica*, the carotenoid content fell by 21.60%, 25.04%, and 27.80%, respectively, at three Cr (VI) concentrations (5, 10, and 15 mg/kg) and by 10.92%, 17.16%, and 19.67% in *S. perspolitana*, compared to the controls. Thus, the organic compounds used in this study indicated a positive effect on the carotenoid content of the two species. Carotenoid content rose by 28.19%, 15.20%, and 11.89% in *S. persica* and 24.52%, 22.18%, and 21.8% in *S. perspolitana* after biochar treatment at Cr (VI) concentrations of 5, 10, and 15 mg/kg, respectively, compared to controls. In addition, in manure treatment at Cr concentrations of 5, 10, and 15 mg/kg, carotenoid content increased by 2.69%, 5.43%, and 8.29% in *S. persica* and 13.02%, 9.99%, and 8.95% in *S. perspolitana*, respectively, compared to controls.

Figure 5 shows the effects of different concentrations of Cr (VI), manure, and biochar on the total phenolic content in *S. persica* and *S. perspolitana*. According to these results, different concentrations of Cr (VI) negatively affected the total phenolic content of the two studied species and so the total phenolic content in the plant decreased with increasing the concentration of Cr in the soil. Total phenolic content in *S. persica* species fell by 39.30%, 39.86%, and 49.73% and in *S. perspolitana* by 34.75%, 42.44%, and 60.35%, respectively, at three Cr (VI) concentrations of 5, 10, and 15 mg/kg. The organic chemicals utilized in this investigation had a good influence on the two species' total phenolic content once again. Total phenolic content raised by 74.83%, 67.59%, and 96.87% in *S. persica* species and by 24.26%, 11.31%, and 36.40% in *S. perspolitana*, respectively, and in biochar treatment at Cr (VI) doses of 5, 10, and 15 mg/kg. In addition, when manure was applied at concentrations of 5, 10, and 15 mg/kg, total phenolic content increased by 39.14%, 33.06%, and 0.052% for *S. persica* and 3.54%, 5.26%, and 22.35% for *S. perspolitana*, respectively, compared to controls.

Determination of TF, BCF, and BAC indices

The results concerning the effect of the studied treatments on the accumulation of metals in Fig. 6 showed that the value of TF for heavy metals was lower than 1 in all treatments. Therefore, both *S. persica* and *S. perspolitana* would serve as suitable species for phytostabilization. The amount of BAC and BCF rose as the concentration of Cr (VI) in the soil increased. Biochar and manure, on the other hand, lowered the BAC and BCF values. At 15 mg/kg of Cr (VI) in the soil without additives, *S. persica* had the higher of these two parameters.

Discussion

Effect of Cr (VI) on germination traits

The current study demonstrated that varying concentrations (5, 25, 100, and 150 mg/kg) of Cr (VI) affected seed germination and seedling growth. In comparison to the control treatment, results with varying doses of Cr (VI) showed a decrease in germination parameters for both *S. persica* and *S. perspolitana* species. This is supported by other research. For example, Cr treatment at 100 mg/kg negatively influenced on germination indices of okra (*Hibiscus esculentus* L.)(Amin et al. 2013). Also, Murtaza et al. (2018) found that the *Vigna radiata* (L.) did not tolerate Cr concentrations higher than 50 mg/L. Besides, both *Salicornia* species indicated decreased growth with increasing Cr (VI) concentration, and it was also found that Cr negatively influenced root and stem length. Bhardwaj et al. (2009) and Peralta et al. (2001) observed similar results for bean and alfalfa plants under cadmium stress. Heavy elements are limiting factors in the germination and growth stages of the seedling. Response to environmental stresses is a complex and undeniable phenomenon in higher plants (Dı́az et al. 2001). Decreased growth in a plant may be due to reduced growth in the roots and consequently deficient transfer of nutrients to the upper parts of the plant. Also, the transfer of Cr to the plant shoot could directly affect its cellular metabolism, which leads to a decreased seedling length (Shanker et al. 2005).

Effect of experimental treatments on plant growth

This study found that varying concentrations of Cr in the soil harmed growth indices in both species, resulting in a drop in plant height as the concentration of Cr in the soil increased, similar to the findings of earlier studies. For example, the presence of Cr (VI) at a concentration of 500 mg/kg in the soil caused a reduction in plant height in *Eruca sativa* species (Kamran et al. 2017). Lukina et al. (2016) found that Cr (VI) (1000 mg/kg) poisoning had a damaging effect on 94% of the 32 plants examined. Besides, reduced root growth and development in the presence of Cr (VI) can result in lower water and nutrient transport to the aerial regions of the plant, resulting in decreased growth and shoot height. Increased Cr (VI) transport to the shoot can also have a direct impact on a leaf, photosynthesis, and cellular metabolism, resulting in plant height reduction (Shahid et al. 2017). Moreover, Cr (VI) has been shown to have a deleterious effect on root dry weight due to root cell destruction. These findings were supported by other scientific literature. Cr (VI)

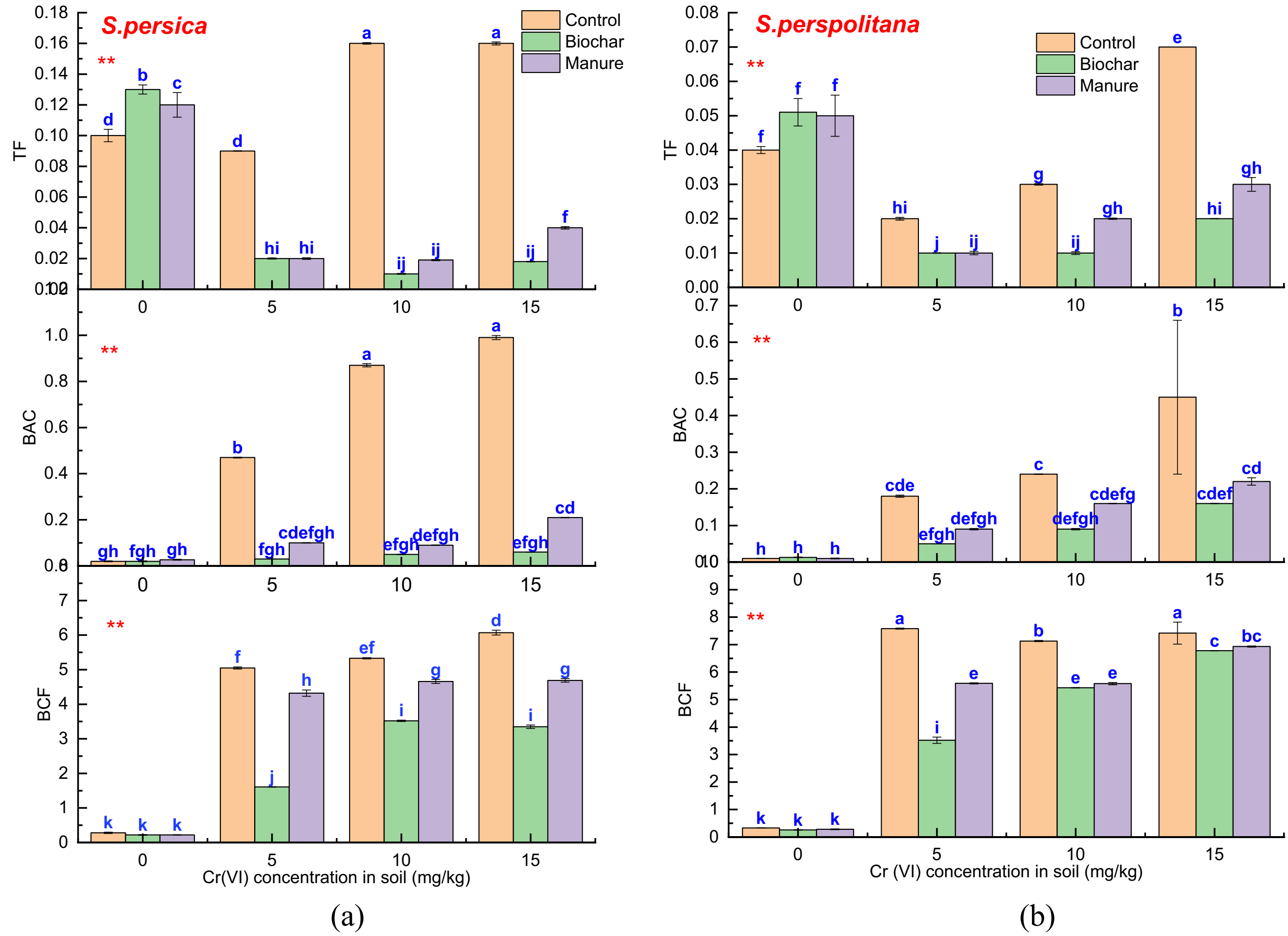

Fig. 6 TF, BCF, and BAC indices for evaluating the ability of phytoremediation of *S. persica* and *S. perspolitana* species grown in contaminated soil with different concentrations of Cr (VI). Error bars represent the standard errors of the means. $p > 0.05$. Asterisk denotes the significance of each factor's interaction effect (species × amendment × concentration); $^{**}p < 0.01$

concentrations greater than 20 and 40 mg/L inhibited plant growth, according to Sundaramoorthy et al. (2010). Shorter roots decreased dry weight, and darker color was observed in *Z. maize* L. grown under Cr (VI) stress (Bhalerao & Sharma 2015).

Our findings also revealed that the applied amendments (manure and biochar) had a positive impact on both species' growth indices (Table 2). This result is in line with the findings of other investigations. Ahmad et al. (2014) investigated the impact of biochar on maize (*Z. mays L.*) biomass and discovered that it rose significantly when compared to the control.

Several variables could be responsible for the increase in shoot and root biomass: (1) biochar's contribution to primary nutrients (e.g., nitrogen) (Meier et al. 2017; Schulz et al. 2013), (2) increase in soil total organic carbon content (Schulz et al. 2013), and (3) decrease of Cr toxicity by immobilization (Chen et al. 2021; Mensah et al. 2022). Adding biochar to the soil, according to Lehmann (2009), maintains nutrients and thus boosts plant growth. Xiang et al. (2017) also stated that biochar could increase crop yield by improving soil nutrients and physical and chemical properties because of its nutrients, porous structure, specific surface, and strong functional groups. Besides, manure's contribution to soil fertility may explain why organic manure has a positive impact on plant height. When manure decomposes, it improves macro and micronutrients and the physico-chemical characteristics of the soil (Chen et al. 2021; Mensah et al. 2022; Moameri et al. 2017; Tiamiyu et al. 2012). Furthermore, manure reduced Cr (VI) mobility in the soil, alleviating Cr (VI)-induced plant stress (VI) (Chen et al. 2021).

The effect of manure, biochar, and Cr (VI) concentration on metal accumulation

The accumulation of Cr (VI) in the roots and shoots of both species rose as the Cr (VI) concentration in the soil increased, according to the results. The majority of the Cr (VI) accumulated in the roots, consistent with Shahid et al. (2017) findings. The Cr content in different portions of *Leersia hexandra Swartz* was as follows: root > stem > leaf > seed. The largest concentration of Cr was found in the root cell wall and intercellular spaces of the rhizome, according to Liu et al. (2009). The synthesis of insoluble Cr compounds in the plant is thought to cause Cr fixation in plant roots. Besides, Cr accumulation in root cells may be increased due to its storage in root cell vacuoles, which could be a plant response to heavy metal toxicity to reduce the heavy metals' harmful potential (Shahid et al. 2017). According to Brunetti et al. (2011), a more significant accumulation of Cr, copper, lead, and zinc in the roots than in the shoots is a method of plant tolerance and adaptation to high metal concentrations in the soil. In addition, Singh et al. (2004) stated that the more considerable accumulation of heavy metals in the roots than in the shoot was attributed to the complexation of these metals with sulfhydryl groups, which prevented the metals from being transferred to the shoot. This prevents heavy metals from entering the food chain. However, although the amount of Cr transferred from plant roots to shoots is low, the amount of Cr transferred inside plant tissue is dependent on its chemical form. The decreased Cr transport to plant shoots could be attributed to the conversion of Cr (VI) to Cr (III) within plants due to Cr (III) tendency's to adhere to cell walls.

The findings of this study revealed that the amendments (biochar and manure) hurt the accumulation of Cr (VI) in the roots and shoots of both species (Figs. 3 and 4). The highest decrease in Cr (VI) uptake was observed in biochar treatment. Adding biochar as an amendment to soils contaminated with heavy metals reduced the availability and absorption of these elements in plants (Namgay et al. 2010). According to Xu et al. (2020), the proportion of –CO in biochar decreased after reaction with Cr (VI), whereas the amount of –C = = O and –COO increased. They concluded that -C–O was engaged in Cr (VI) reduction and oxidized to –C = = O and –COO subsequently. Furthermore, adding manure to a soil that includes much humus may alter the availability of heavy metals by changing the physical and chemical properties of the soil (e.g., enhancing the cation exchange capacity) (Pinto et al. 2004). In other words, the availability and mobility of Cr (VI) in soil were reduced, which resulted in a decrease in Cr (VI) uptake. Besides, on the surface of the manure, oxygen-containing functional groups like -OH could form complexes with $HCrO_4$ and $Cr_2O_7^{2-}$, reducing Cr (VI) migration (Chen et al. 2021; Zhang et al. 2017). For example, Antoniadis et al. (2017) demonstrated that adding manure to *Cichorium spinosum* L. reduces the bioavailability of Cr. Chen et al. (2021) also found that chicken manure significantly reduced Cr (VI) uptake by *Pharbitis purpurea*.

Effect of manure, biochar, Cr (VI) concentrations on chlorophyll a and b, carotenoid, and phenolic content

The results of the present study showed that total phenolic content and photosynthetic pigments significantly decreased because of Cr (VI) toxicity. As shown in Fig. 5, this decrease was more remarkable in *S. perspolitana* than in *S. persica* and probably due to the higher Cr (VI) accumulation in *S. perspolitana* (Figs. 3 and 4). This was in line with the findings of Zeeshan et al. (2020). They found that tomato plants showed declines in chlorophyll a and b, total chlorophyll, and carotene when exposed to Ni, Pb, and Cd pollution. Similar findings were also reported for maize (*Z. mays* L.) (Rahmaty &Khara 2011) and wheat (*Triticum aestivum* L.) (Subrahmanyam 2008). Moreover, different Cr (VI) concentrations had a substantial impact on the plant's phenolic content, to the point where an accumulation of Cr (VI) at a concentration of 15 mg/kg lowered the plant's antioxidant traits. This agrees with Levizou et al. (2019b)'s findings. They showed that the total phenolic content in the *Origanum vulgare* L. decreased with the increasing Cr in the soil. Cr causes toxicity in plants by interfering with many metabolic pathways. A decrease in the activity of the d-aminolevulinic acid dehydratase has been connected to a reduction in chlorophyll production owing to metal stress (e.g., Cr (VI)) (Oliveira 2012, Prasad &Prasad 1987, Vajpayee et al. 2000). In addition, the organic compounds (biochar and manure) increased in the number of photosynthetic pigments, with biochar having a more significant influence than manure (Fig. 5). It can be explained by the potential of the amendments to stimulate plant development by improving soil physio-chemical characteristics and nutrient availability while also lowering heavy metal accumulation (Figs. 3 and 4). Lower Cr (VI) accumulation in *Salicornia* plants were observed where the biochar was amended. Besides, biochar improved photosynthetic function in leaves by assuring the synthesis of a number of enzymes and electron transporters essential in photosynthesis (Hou et al. 2021).

Determination of TF[1], BCF[2], and BAC[3] indices

The results of phytoremediation in the present study showed that *S. persica* and *S. perspolitana* could absorb heavy metals from the soil with higher uptake of metals in the roots than shoots for both species. Considering that the TF index was lower than 1 for all treatments, one can conclude that *S. persica* and *S. perspolitana* were not suitable for phytoextraction of metal Cr. Also, the BCF factor for Cr (VI) was higher than 1, indicating that both *S. persica* and *S. perspolitana* stabilize the metal in the soil during the stabilization process. Both species mostly absorb the trivalent form of Cr

(VI), indicating the high efficiency for converting hexavalent to trivalent form.

Yoon et al. (2006) found that plants with high accumulation and low transfer factors can stabilize heavy metals. Plant species that have been used to stabilize heavy metals so far include Festuca rubra L. (lead and zinc) and Brassica juncea (L.) (cadmium) (Ghosh &Singh 2005), which is consistent with our findings in this research.

The use of soil amendments in phytoremediation aims to improve the efficiency of the cleaning process. Depending on the needs, organic amendments can be used to encapsulate pollutants or stimulate their uptake and transfer to harvestable plant biomass (Wiszniewska et al. 2016). TF, BCF, and BAC were reduced in this study by adding biochar and manure to contaminated soil (Fig. 6). This suggests that these two compounds decreased Cr (VI) bioavailability in the soil and prevented the metal from entering the plant's roots and shoots, resulting in higher plant growth and establishment in polluted soil (Table 2). Organic amendments increased phytoremediation efficacy by lowering plant absorption and increasing Cr (VI) fixation in the soil, which is important because Cr (VI) is extremely mobile and toxic in soil.

Cr (VI) concentrations in the edible part (stem) of S. persica and S. perspolitana growing in polluted soil ranged from 0.65 to 1.93 mg/kg and 0.37 to 1.98 mg/kg, respectively. The concentration of Cr (VI) in both species exceeded the maximum permissible limits of the World Health Organization (WHO)/Food Agriculture Organization (FAO) (WHO 2000) for plants (0.10 mg/kg) in the contaminated soil. The content of Cr (VI) in the edible part of S. persica was reduced by 23 to 48% when biochar was added to the soil as an amendment, whereas the reduction for S. perspolitana was 24 to 61%. Manure also lowered Cr (VI) levels in S. persica stems by 16 to 45% and in S. perspolitana stems by 10 to 50%, respectively. Although these two amendments significantly decreased the concentration of Cr (VI) in the edible section of Salicornia and thus reduced the possible health risks of chromium, their levels were still above the FAO maximum permissible limit, indicating that consumers were not safe.

Conclusions

The ability of *S. persica* and *S. perspolitana* to accumulate Cr (VI) was investigated in the presence of manure and biochar as a soil supplement. Cr (VI) accumulation in the shoots and roots of the two species rose as the metal concentration in the soil increased. *S. persica* accumulated higher total Cr, Cr (III), and Cr (VI) in its shoots than *S. perspolitana*. With rising Cr concentrations, photosynthetic pigments and total phenolic content in both tested species declined. Adding biochar and manure to the soil stimulated the growth of *S. persica* and *S. perspolitana* while lowering the Cr concentration in their roots and shoots. Biochar treatment reduced heavy metal accumulation in plant tissues significantly more than manure treatment. Both *S. persica* and *S. perspolitana* can phytostabilize Cr (VI) because the BCF factor is higher than one. It can be concluded that utilizing organic amendments in heavy metal-contaminated soils can help to stabilize the metals and keep them out of the food chain.

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Author contribution Fahime Ashrafi: investigation, project administration, resources, validation, writing–original draft, software, formal analysis, and funding acquisition.

Ava Heidari: supervision, visualization, writing–review and editing, conceptualization, data curation, methodology, project administration, and funding acquisition

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Declarations

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